OCT 1 8 2011

510(k) SUMMARY

This summary of 510(k) safety and effectiveness information is being submitted in accordance with the requirements of SMDA 1990 and 21 CFR 807.92.

The assigned 510(k) number is **K111904**.

807.92 (a)(1): Name: **ARK Diagnostics, Inc.**

Address: 1190 Bordeaux Drive
Sunnyvale, CA 94089

Owner Operator Number: 10027663
Establishment Registration: 3005755244

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Executive Director of Quality and Regulatory Affairs

Date prepared: September 7, 2011

807.92 (a)(2): Device name- trade name and common name, and classification

Trade name: ARK™ Methotrexate Assay
ARK™ Methotrexate Calibrator
ARK™ Methotrexate Control

Common Name: Homogeneous Enzyme Immunoassay

Classification: 21 CFR 862 Clinical Chemistry Test System - Toxicology; Test Code LAO; Enzyme Immunoassay, Methotrexate Pre-Amendment Device, Unclassified
(21 CFR 862.3200 DLJ, 21 CFR 862.3280 LAS)

807.92 (a)(3): Identification of the legally marketed predicate device

Abbott TDx®/TDxFLx® METHOTREXATE II (K932615)

807.92 (a)(4): Device Description

The ARK Methotrexate Assay is a homogeneous immunoassay based on competition between drug in the specimen and Methotrexate labeled with the enzyme glucose-6-phosphate dehydrogenase (G6PDH) for binding to the antibody reagent. As the latter binds antibody, enzyme activity decreases. In the presence of drug from the specimen, enzyme activity increases and is directly proportional to the drug concentration. Active enzyme converts the coenzyme nicotinamide adenine dinucleotide (NAD) to NADH that is measured spectrophotometrically as a rate of change in absorbance. Endogenous serum G6PDH does not interfere with the results because the coenyzme NAD functions only with the bacterial enzyme used in the assay.

The ARK Methotrexate Assay consists of reagents R1 anti-Methotrexate polyclonal antibody with substrate and R2 Methotrexate labeled with bacterial G6PDH enzyme. The ARK Methotrexate Calibrator consists of a six-level set to calibrate the assay, and the ARK Methotrexate Control consists of a six-level set used for quality control of the assay (tri-level calibration range set and tri-level high range set). The ARK Methotrexate Dilution Buffer is equivalent to zero calibrator (Calibrator A).

807.92 (a)(5): Intended Use / Indications for Use

The ARK Methotrexate Assay is a homogeneous enzyme immunoassay intended for the quantitative determination of methotrexate in human serum or plasma on automated clinical chemistry analyzers. The measurements obtained are used in monitoring levels of methotrexate to help ensure appropriate therapy.

The ARK Methotrexate Calibrator is intended for the calibration of the ARK Methotrexate Assay.

The ARK Methotrexate Control is intended for the quality control of the ARK Methotrexate Assay.

Specimens from patients who have received glucarpidase (carboxypeptidase G2) as a high dose methotrexate rescue therapy should ***not*** be tested with the ARK Methotrexate Assay.

807.92 (a)(6): Technological Similarities and Differences to the Predicate

SUBSTANTIAL EQUIVALENCE COMPARATIVE CHART

Comparison between the ARK™ Methotrexate Assay and TDx®/TDxFLx® Methotrexate II Assay

Characteristic	Device ARK™ Methotrexate Assay	Predicate Abbott TDx®/TDxFLx® Methotrexate II (K932615)
Intended Use	The ARK™ Methotrexate Assay is intended for the quantitative determination of methotrexate in human serum or plasma on automated clinical chemistry analyzers.	The TDx®/TDxFLx® Methotrexate II assay is a reagent system for the quantitative measurement of methotrexate, an antineoplastic drug, in serum or plasma.
Indications for Use	The measurements obtained are used in monitoring levels of methotrexate to help ensure appropriate therapy.	The measurements obtained are used in monitoring levels of methotrexate to ensure appropriate therapy.
Sample	Serum or plasma	Serum or plasma
Methodology	Homogenous enzyme immunoassay (EIA)	Fluorescence polarization immunoassay (FPIA)
Reagent Components	Two (2) reagent system: Anti-Methotrexate Antibody/Substrate Reagent (R1) containing rabbit polyclonal antibodies to Methotrexate, glucose-6-phosphate, nicotinamide adenine dinucleotide, bovine serum albumin, preservatives, and stabilizers Enzyme Reagent (R2) containing Methotrexate labeled with bacterial G6PDH, buffer, bovine serum albumin, preservatives, and stabilizers	Reagent Pack: W = Wash solution, solvent, sodium azide S = Methotrexate Antibody (mouse monoclonal), buffer, protein and sodium azide T = Methotrexate Fluorescein Tracer, buffer, protein, surfactant and sodium azide P = Pretreatment Solution, protein, surfactant and sodium azide
Platform required	Automated clinical chemistry analyzer	TDx clinical chemistry analyzer
Accessory reagents	Calibrators (six levels) and controls (six levels) in a synthetic matrix; Dilution Buffer	Calibrators (six levels) and controls (six levels) in human serum; Dilution Buffer
Testing environment	Routine clinical laboratory	Routine clinical laboratory
Reagent condition and storage	Liquid, 2-8° C	Liquid, 2-8° C

807.92 (b)(1) and 807.92 (b)(2):
Brief Description of Nonclinical and Clinical Data

Limit of Quantitation (LOQ)

The following characteristics were determined according to CLSI EP17-A for the ARK Methotrexate Assay. Analyzer-specific performance may vary.

Criterion	MTX Concentration (µmol/L)
Limit of Blank (LoB); N = 60 µB + 1.645 SD , where SD = 0.005	0.01
Limit of Detection (LoD); N = 60 LoB + 1.652 SD, where SD = 0.005	0.02
Limit of Quantitation (LoQ); N = 40 LoQ – 2 SD > LoD	0.04

Each laboratory is responsible for determining reporting criteria for methotrexate concentrations. The following suggestion from CLSI EP17-A may be appropriate:

Result ≤ LoB	report "not detected; concentration < LoD"
LoB < Result < LoQ	report "analyte detected; concentration < LoQ"
Result ≥ LoQ	report the result as measured

Accuracy

Accuracy (analytical recovery) was performed by adding concentrated methotrexate drug into human serum negative for methotrexate. A certified stock concentrate of highly pure methotrexate was added volumetrically to human serum negative for methotrexate, representing drug concentrations across the assay calibration range. Six replicates of each sample were assayed on an automated clinical chemistry analyzer. The results were averaged and compared to the target concentration and percent recovery calculated. Results are shown below.

$$\% \text{ Recovery} = 100 \times \frac{\text{Mean recovered concentration}}{\text{Target concentration}}$$

Theoretical Concentration (µmol/L)	Mean Recovered Concentration (µmol/L)	Percentage Recovery
0.06	0.07	111.1
0.10	0.10	100.0
0.30	0.30	98.3
0.60	0.61	102.2
1.00	0.99	98.8

Mean percentage recovery: 102.1

Linearity

Linearity studies were performed as suggested in CLSI/NCCLS Protocol EP6-A. A 1.30 µmol/L serum sample was prepared and dilutions were made proportionally with human serum negative for methotrexate. Linearity at specific dilutions was considered acceptable if the percent difference was ±10% between the predicted 1st and 2nd order regressed values for concentrations >0:10 µmol/L or ±0.01 µmol/L at concentrations ≤ 0.10 µmol/L. Results are shown below.

Theoretical (µmol/L)	Observed Results (µmol/L)	1st Order Predicted Results	2nd Order Predicted Results	Difference (µmol/L or %)
0.00	0.00	0.009	-0.003	na
0.02	0.02	0.026	0.016	-0.010 µmol/L
0.04	0.04	0.042	0.034	-0.008 µmol/L
0.05	0.06	0.059	0.053	-0.006 µmol/L
0.07	0.08	0.076	0.072	-0.004 µmol/L
0.11	0.11	0.110	0.109	-0.7 %
0.18	0.17	0.178	0.183	3.1 %
0.36	0.34	0.347	0.364	4.8 %
0.65	0.63	0.618	0.639	3.4 %
0.72	0.72	0.686	0.705	2.9 %
0.86	0.84	0.821	0.835	1.7 %
1.01	0.99	0.957	0.960	0.4 %
1.15	1.06	1.092	1.082	-1.0 %
1.30*	1.19	1.228	1.199	-2.3 %

*Concentration exceeds the claimed calibration range.

Samples containing methotrexate between 2 and 1200 µmol/L were prepared proportionally in pooled human serum and then diluted into the calibration range with ARK Methotrexate Dilution Buffer. Regression of assayed methotrexate concentrations was linear throughout the range.

Assay Range

The measurement range of the ARK Methotrexate Assay is 0.04 - 1.20 µmol/L. Specimens containing methotrexate in higher concentrations are assayed by dilution of the specimen. Report assayed values exceeding the LoD according to the information provided for LoQ. Multiply the assayed result by the dilution factor for specimens containing methotrexate above the measurement range.

Method Comparison

Correlation studies were performed using CLSI/NCCLS Protocol EP9-A2. Results from the ARK Methotrexate Assay were compared with results from Fluorescence Polarization Immunoassay method (monoclonal FPIA).

Methotrexate concentrations by FPIA ranged 0.04 to 1440 µmol/L (µM). ARK Methotrexate values ranged 0.04 to 1500 µmol/L. Results of the Passing-Bablok regression analysis for the study are shown below (with 95% confidence limits) for 102 specimens within the measurement range as well as for all 147 specimens including those above the measurement range requiring dilution.

Parameter	**Range 0.04 to 1.19 µM**		**Range 0.04 to 1440 µM**	
Slope	1.00	(1.00 to 1.02)	0.99	(0.96 to 1.00)
y-intercept	0.01	(0.00 to 0.01)	0.01	(0.01 to 0.01)
Correlation Coefficient (r^2)	0.978	(0.968 to 0.985)	0.998	(0.997 to 0.998)
Number of Samples	102	na	147	na

Precision

Precision was determined as described in CLSI/NCCLS Protocol EP5-A2. The six-level ARK Methotrexate Control and pooled human specimens containing methotrexate were used in the study. Each level was assayed in quadruplicate twice a day for 20 days. Each of the runs per day was separated by at least two hours. The within-run, between-day, total SD, and percent CVs were calculated. Results are shown below. Acceptance criteria: ≤10% total CV at >0.10 µmol/L, SD ≤0.01 at ≤0.10 µmol/L.

Sample	N	Mean (µmol/L)	Within Run		Between Day		Total	
			SD	%CV	SD	%CV	SD	%CV
ARK Methotrexate Control								
LOW	160	0.06	0.005	8.1	0.005	7.1	0.007	10.6
MID	160	0.37	0.011	3.1	0.008	2.2	0.014	3.8
HIGH	160	0.76	0.039	5.1	0.029	3.8	0.048	6.4
5	160	4.8	0.13	2.8	0.013	2.8	0.19	4.1
50	160	48	1.40	2.9	2.13	4.4	2.71	5.6
500	160	470	15.63	3.3	27.64	5.8	33.35	7.0
Patient Pool								
LOW	160	0.07	0.006	9.1	0.005	7.5	0.008	11.7
MID	160	0.41	0.013	3.3	0.026	6.4	0.030	7.2
HIGH	160	0.82	0.037	4.5	0.042	5.1	0.057	6.9
5	160	4.6	0.14	3.1	0.18	4.0	0.24	5.3
50	160	45	1.31	2.9	2.62	5.9	2.92	6.5
500	160	460	11.55	2.5	27.21	5.9	29.63	6.4

Interfering Substances

Interference studies were conducted using CLSI/NCCLS Protocol EP7-A2 as a guideline. Clinically high concentrations of the following potentially interfering endogenous substances in serum with known levels of methotrexate (approximately 0.05 and 0.50 µmol/L) were evaluated. Each sample was assayed using the ARK Methotrexate Assay, along with a serum control of methotrexate. Measurement of methotrexate was not substantially affected at the levels of endogenous substances tested.

		Methotrexate (~ 0.05 µmol/L)		Methotrexate (~ 0.50 µmol/L)	
Interfering Substance	**Interferent Concentration**	**Serum Control**	**Test**	**Serum Control**	**Test (% Control)**
Albumin	12 g/dL	0.05	0.06	0.48	0.45 (92.8)
Bilirubin - conjugated	70 mg/dL	0.05	0.06	0.48	0.51 (105.5)
Bilirubin - unconjugated	70 mg/dL	0.05	0.06	0.48	0.52 (106.9)
Cholesterol	400 mg/dL	0.05	0.06	0.47	0.49 (105.4)
Gamma-Globulin	12 g/dL	0.05	0.06	0.48	0.51 (105.5)
Hemoglobin	1000 mg/dL	0.04	0.05	0.49	0.45 (92.8)
Intralipid®	500 mg/dL	0.05	0.05	0.43	0.45 (105.1)
Rheumatoid Factor	1100 IU/mL	0.05	0.06	0.43	0.41 (96.1)
Triglycerides	749 mg/dL	0.04	0.04	0.49	0.45 (91.4)
Uric Acid	30 mg/dL	0.05	0.04	0.48	0.50 (102.8)

Specificity - Drug Interference

Crossreactivity to 7-Hydroxymethotrexate, the major metabolite

The ARK Methotrexate Assay did not crossreact (≤ 0.07%) with the major metabolite 7-Hydroxymethotrexate.

Crossreactivity to the minor, inactive metabolite 2,4-diamino-N^{10}-methylpteroic acid (DAMPA)

The ARK Methotrexate Assay crossreacts substantially with the minor metabolite DAMPA. Tests were performed in the absence of the parent drug methotrexate. Crossreactivity to DAMPA ranged 64.3 to 100%. The assay should not be used during possible compassionate therapy with glucarpidase (carboxypeptidase G2) that rapidly converts circulating methotrexate to DAMPA.

Drugs that crossreact

The ARK Methotrexate Assay crossreacts slightly with triamterene and trimethoprim, however these drugs may be contraindicated for methotrexate cancer treatement due to additional adverse effects if co-administered. The structures of these compounds closely match the pteridine ring moiety of methotrexate.

Compound	Tested (µmol/L)	Methotrexate Absent MTX (µmol/L)	Methotrexate Absent Cross Reactivity (%)	Methotrexate Present 0.05 µmol/L MTX (µmol/L)	Methotrexate Present 0.05 µmol/L Cross Reactivity (%)	Methotrexate Present 0.50 µmol/L MTX (µmol/L)	Methotrexate Present 0.50 µmol/L Cross Reactivity (%)
Triamterene	25	0.46	1.85	0.89	3.32	1.04	2.31
Trimethoprim	100	0.17	0.17	0.16	0.12	0.99	0.54

Crossreactivity to folate analogs and other compounds

The ARK Methotrexate Assay did not crossreact (≤ 0.01%) with folate analogs or other compounds at ≥ 1000 µmol/L as tested.

Compound	Tested (µmol/L)
Adriamycin	1000
Cyclophosphamide	1500
Cytosine	1000
Dihydrofolic Acid	1000
DL-6-Methyl-5,6,7,8-Tetrahydropterine	1000
Folic Acid	1000
Folinic Acid (leucovorin)	1000
5-Fluorouracil	3000
6-Mercaptopurine	1000
5-Methyltetrahydrofolic acid	1000
Prednisolone	1000
Pyrimethamine	1000
Sulfamethoxazole	1600
Tetrahydrofolic Acid	1000
Vinblastine	1000
Vincristine	1000

Anticoagulants

Studies were conducted to determine the performance characteristics of the assay for both serum and plasma samples containing methotrexate.

The results indicate that there is no significant difference between the recovery of methotrexate in serum or plasma.

Sample Stability

Human specimens were shown to be stable frozen (at least 15 months), forty-eight (48) hours at room temperature, refrigerated (at least 21 days) and after three (3) successive freeze/thaw cycles.

On-Board Stability

Calibration Curve Stability: A stored calibration was effective up to at least 19 days based on supporting data.

Reagent on-board stability: Reagents were effective when stored after transfer to analyzer specific reagent containers for up to at least 25 days based on supporting data. In-use stability of calibrator and controls was also demonstrated.

807.92 (b)(3): Conclusions from Nonclinical Testing

As summarized above, the ARK Methotrexate Assay System, including the ARK Methotrexate Calibrator, ARK Methotrexate Control and ARK Methotrexate Dilution Buffer, is substantially equivalent to the Abbott TDx®/TDxFLx® METHOTREXATE II Assay system. The ARK Methotrexate Assay system was shown to be safe and effective for its intended use based on performance studies.

DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food and Drug Administration
10903 New Hampshire Avenue
Silver Spring, MD 20993

OCT 1 8 2011

ARK Diagnostics, Inc
c/o Kenneth C. Kasper, PhD
1190 Bordeaux Dr.
Sunnyvale, CA 94089

Re: k111904
Trade/Device Name: ARK™ Methotrexate Assay, ARK™ Methotrexate Calibrator, ARK™ Methotrexate Control
Regulatory Class: Unclassified, 510(k) required
Product Code: LAO, DLJ, LAS
Dated: September 7, 2011
Received: September 8, 2011

Dear Dr. Kasper:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

If your device is classified (see above) into class II (Special Controls), it may be subject to such additional controls. Existing major regulations affecting your device can be found in Title 21, Code of Federal Regulations (CFR), Parts 800 to 895. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Parts 801 and 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR 803); and good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820). This letter will allow you to begin marketing your device as described in your Section 510(k) premarket notification. The FDA finding of substantial equivalence of your device to a legally marketed predicate device results in a classification for your device and thus, permits your device to proceed to the market.

If you desire specific advice for your device on our labeling regulation (21 CFR Part 801), please contact the Office of *In Vitro* Diagnostic Device Evaluation and Safety at (301) 796-5450. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21 CFR Part 807.97). For questions regarding postmarket surveillance, please contact CDRH's Office of Surveillance and Biometric's (OSB's) Division of Postmarket Surveillance at (301) 796-5760. For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to http://www.fda.gov/MedicalDevices/Safety/ReportaProblem/default.htm for the CDRH's Office of Surveillance and Biometrics/Division of Postmarket Surveillance.

You may obtain other general information on your responsibilities under the Act from the Division of Small Manufacturers, International and Consumer Assistance at its toll-free number (800) 638-2041 or (301) 796-5680 or at its Internet address http://www.fda.gov/MedicalDevices/ResourcesforYou/Industry/default.htm.

Sincerely yours,

Courtney H. Lias, Ph.D.
Director
Division of Chemistry and Toxicology
Office of *In Vitro* Diagnostic Device
Evaluation and Safety
Center for Devices and Radiological Health

Enclosure

Indication for Use

510(K) Number (if known): K111904

Device Name:

ARK™ Methotrexate Assay
ARK™ Methotrexate Calibrator
ARK™ Methotrexate Control

Indications for Use:

The ARK™ Methotrexate Assay is a homogeneous enzyme immunoassay intended for the quantitative determination of Methotrexate in human serum or plasma on automated clinical chemistry analyzers. The measurements obtained are used in monitoring levels of methotrexate to help ensure appropriate therapy.

The ARK™ Methotrexate Calibrator is intended for use in calibration of the ARK Methotrexate Assay.

The ARK™ Methotrexate Control is intended for use in quality control of the ARK Methotrexate Assay.

Specimens from patients who have received glucarpidase (carboxypeptidase G2) as a high dose methotrexate rescue therapy should ***not*** be tested with the ARK Methotrexate Assay.

Prescription Use ___X___ (21 CFR Part 801 Subpart D)	And/Or	Over the Counter Use ____. (21 CFR Part 801 Subpart C)

(PLEASE DO NOT WRITE BELOW THIS LINE; CONTINUE ON ANOTHER PAGE IF NEEDED)

Concurrence of CDRH, Office of In Vitro Diagnostic Device Evaluation and Safety (OIVD)

Ruth Chesler

Division Sign-Off
Office of In Vitro Diagnostic Device
Evaluation and Safety

510(k) 111904